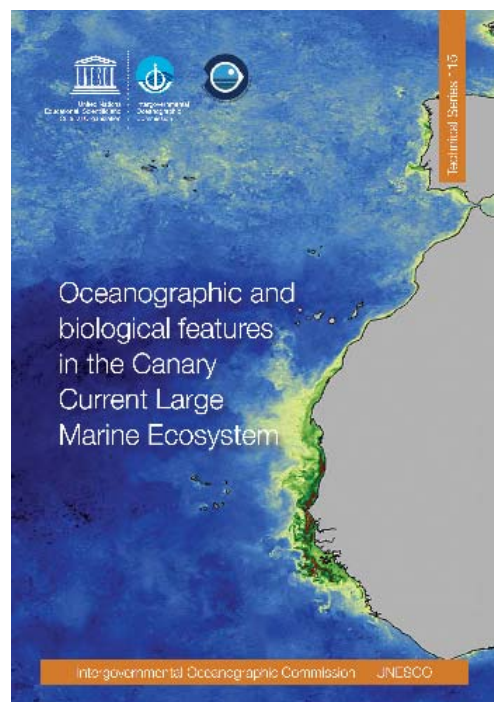


For bibliographic purposes, this separate part should be cited as:

Pitcher, G. C. and Fraga, S. 2015. Harmful algal bloom events in the Canary Current Large Marine Ecosystem. In: Valdés, L. and Déniz-González, I. (eds). *Oceanographic and biological features in the Canary Current Large Marine Ecosystem*. IOC-UNESCO, Paris. IOC Technical Series, No. 115, pp. 175-182. doi: XXXXXXXXXX

The publication should be cited as follows:

Valdés, L. and Déniz-González, I. (eds). 2015. *Oceanographic and biological features in the Canary Current Large Marine Ecosystem*. IOC-UNESCO, Paris. IOC Technical Series, No. 115: 383 pp.



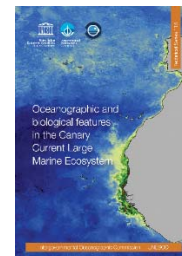
The report *Oceanographic and biological features in the Canary Current Large Marine Ecosystem* and its separate parts are available on-line at: <http://www.unesco.org/new/en/ioc/ts115>.

The bibliography of the entire publication is listed in alphabetical order in pages 351-379. The bibliography cited in this article was extracted and listed in alphabetical order at the end of this separate part, in unnumbered pages.

ABSTRACT

We provide a review of Harmful Algal Blooms (HABs) within the Canary Current Large Marine Ecosystem (CCLME). As yet all documented HABs within the region have been associated with the production of one or another toxin. The diversity of harmful algae recorded within the region is similar to that found in other eastern boundary upwelling systems, and includes those species responsible for paralytic shellfish poisoning, diarrhetic shellfish poisoning, amnesic shellfish poisoning and azaspiracid poisoning. Also present off Northwest Africa, but generally absent from the other major upwelling systems, are those species responsible for ciguatera fish poisoning and microcystin-producing cyanobacterial blooms. Their presence is afforded by the subtropical habitat provided by the island archipelagos found within the CCLME. It is intended that this brief review will provide the foundation and stimulus for further studies of the ecology and dynamics of HABs, of their toxins, and of the public health and socioeconomic impacts of HABs within this region.

Keywords: *Paralytic shellfish poisoning, Diarrhetic shellfish poisoning, Amnesic shellfish poisoning, Azaspiracid poisoning, Ciguatera fish poisoning, Canary Current Large Marine Ecosystem, Northwest Africa*



HARMFUL ALGAL BLOOM EVENTS IN THE CANARY CURRENT LARGE MARINE ECOSYSTEM

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4.5.1. INTRODUCTION

The major eastern boundary upwelling systems (EBUS) of the world's oceans are susceptible to harmful algal blooms (HABs) because they are highly productive, nutrient-rich environments, prone to high-biomass blooms (Pitcher et al., 2010; Trainer et al., 2010). The majority of species contributing to HABs within these systems constitute regular components of the phytoplankton assemblages of upwelling systems, and their harmful impacts are associated with either their toxic properties or the high biomass such blooms can achieve. Bloom development is dictated through the creation of a mosaic of shifting habitats determined by windstress fluctuations and buoyancy inputs at several scales, and by mesoscale features that interrupt typical upwelling circulation patterns.

As part of an EBUS the Canary Current Large Marine Ecosystem (CCLME) extends from the northwestern tip of Morocco to the south of Guinea-Bissau, and includes the Canary Islands and Cape Verde. The LME is characterized by strong sub-regional variability linked to variable coastline orientation, configuration and shelf width, variable coastal upwelling, nutrient and freshwater input, and retentive or dispersive physical mechanisms (Aristegui et al., 2009). HABs of the CCLME have been poorly studied although estimates of chlorophyll and primary production, as derived from satellite ocean colour data, have been shown by Chavez and Messié (2009) to be high, indicating in all likelihood a high incidence of HABs.

The sub-regions within the CCLME demonstrate latitudinal variability in satellite derived surface chlorophyll concentrations linked to a southward increase in nutrient concentrations (Lathuilière et al., 2008; Aristegui et al., 2009). The northernmost region includes the Moroccan coastline of the Gulf of Cadiz which is unfavourable for upwelling. Further south, between Cape Sim and Cape Blanc, the coasts of Morocco and Western Sahara benefit from year-round upwelling of North Atlantic Central Water (NACW) and are characterized by high mesoscale variability arising from geographical heterogeneity. Here variations in shelf width and the presence of major capes produce extended filaments of elevated chlorophyll. South of Cape Blanc the Mauritanian-Senegalese region is the most productive as a result of winter upwelling of higher-nutrient South Atlantic Central Water (SACW). Also, present within the CCLME are subtropical island archipelagos which span the coastal transition zone from near to open coast providing an additional habitat for HABs. We piece together the few well-documented studies of HABs within these sub-regions to provide a better picture of the incidence and likely causes and impacts of HABs off northwest Africa (NWA). As yet all documented HABs within the region have been associated with the production of one or another toxin.

4.5.2. TOXIN-PRODUCING ALGAE IN THE CCLME

4.5.2.1. Paralytic shellfish poisoning toxins

Paralytic shellfish poisoning (PSP) is associated with the consumption of seafood products contaminated with the neurotoxins collectively known as saxitoxins (STXs) (Etheridge, 2010). Major toxin sources include several species of dinoflagellates of the genera *Alexandrium*, *Gymnodinium* and *Pyrodinium*. Within EBUS *Alexandrium catenella* and *Gymnodinium catenatum* are the most common source of PSP toxins (Trainer et al., 2010). Specific association of *G. catenatum* with the Iberian upwelling system followed a major PSP event on the Galician coast in 1976.

Off NWA records of PSP following consumption of shellfish date back to 1961 on the Moroccan coast with further records in the 1970s and 1980s (Taleb et al., 2003). In 1992 a comprehensive monitoring programme was initiated with the analysis of several species of bivalve molluscs collected fortnightly at 30 stations located on the Atlantic coast of Morocco and off Western Sahara (Taleb et al., 2003). Through implementation of this programme shellfish contaminated with PSP toxins were found in November 1994 to extend from Larache to Essaouira with the highest toxin concentrations of 6000 µg STX eq 100 g⁻¹ in mussels (*Mytilus galloprovincialis*) from the Casablanca region (Taleb et al., 1998, 2003). Maximum toxin concentration in oysters (*Crassostrea gigas*) and clams (*Ruditapes decussatus*) was 2608 and 738 µg STX eq 100 g⁻¹ respectively. At that time the presence of *Gymnodinium catenatum* (Figure 4.5.1a) in the waters around Casablanca (maximum recorded concentration of 3x10⁴ cells l⁻¹) indicated that it was the most likely cause (Tahri Joutei, 1998); an observation supported by the complex toxin profile of the mussels, that included the presence of dcSTX and the absence of STX (Taleb et al., 2003). In November 1995 shellfish contaminated by PSP toxins were again present near Casablanca although levels of toxicity were much lower.

In 1998 and 1999 shellfish contaminated by PSP toxins were also recorded further south between Agadir and Tan Tan (Taleb et al., 2003). A maximum level of PSP toxins of 1142 µg STX eq 100 g⁻¹ was recorded in mussels from Taghazout and the toxin profile of primarily GTX1-4 toxins implicated *Alexandrium minutum* as a possible toxin source; further supported by observations of this species as a component of the plankton (Tahri Joutei et al., 2000).

The prominence of *G. catenatum* within the CCLME has been further demonstrated by its presence in recent sediments of the North Canary Basin (Targarona et al., 1999). Here cysts of *Gymnodinium* species (dominated by *G. catenatum*, Figure 4.5.1b,c), of *Lingulodinium machaerophorum* (= *Lingulodinium polyedrum*), and of *Protoperidinium* species have been found to dominate cyst assemblages occurring in higher numbers in association with the upwelling centers of Cape Ghir and Cape Jubi. These findings support speculation by Ribeiro et al. (2012) that refuge cyst populations off NWA may have been the source of *G. catenatum* blooms off the Iberian coast in the 1970s.

In addition to the threat posed to human health, strong evidence has been provided for PSP toxins as the cause of a mass mortality of Monk Seals in the Cape Blanc peninsula in 1997 (Reyero et al., 1999). The mortality of two-thirds of the population of this endangered species was initially attributed to a morbillivirus (Osterhaus et al., 1997). However, epidemiological, clinical, pathological and toxicological observations were more consistent with poisoning by PSP toxins, and their presence was confirmed in both seal tissue and other fauna (Hernández et al., 1998). Mouse bioassays showed symptoms of PSP, and high performance liquid chromatography (HPLC) supported by mass spectrometry (MS) indicated the presence of dcSTX and neoSTX (Reyero et al., 1999). *G. catenatum* was again considered to be the most likely origin

of these toxins based on the toxin composition of analysed samples and its known presence within the CCLME.



Figure 4.5.1. Scanning electron micrograph of (A) the vegetative stage of the dinoflagellate *Gymnodinium catenatum* (photograph by S. Fraga) the cause of PSP contaminated shellfish on the Moroccan coast (Taleb et al., 2003), and (B and C) light micrographs of the cyst stage of *G. catenatum* (photographs by R. Figueroa) found to dominate recent sediments of the North Canary Basin (Targarona et al., 1999).

4.5.2.2. Diarrhetic shellfish poisoning and other lipophilic toxins

Diarrhetic shellfish poisoning (DSP) is characterized by severe gastrointestinal illness associated with the consumption of filter feeding bivalves (Reguera et al., 2012). Historically three different toxin groups were associated with DSP: okadaic acid (OA) and the dinophysistoxins (DTXs), pectenotoxins (PTXs) and yessotoxins (YTXs) as a result of their co-extraction and single response to the conventional mouse bioassay. However, diarrheagenic effects have only been proven for OA and DTXs, which together with the PTXs are typically produced by species of the dinoflagellate genus *Dinophysis*. Although no longer associated with DSP the YTXs, produced by the dinoflagellates *Protoceratium reticulatum*, *Lingulodinium polyedrum* and *Gonyaulax spinifera*, are lethal to mice following intraperitoneal injection (Tubaro et al., 2010). On the other hand, the more recently discovered azaspiracids (AZAs), produced by the dinoflagellate *Azadinium spinosum*, although never associated with the DSP toxins are known to cause diarrhea in humans (Furey et al., 2010).

DSP toxins, YTXs and the AZAs have all been shown to be present in EBUS and DSP off the Iberian Peninsula, attributed primarily to *D. acuminata*, *D. acuta* and *D. caudata*, is considered to pose the greatest threat to

shellfish harvests in this area (Trainer et al., 2010). Off NWA lipophilic toxins have been recorded almost annually on the Moroccan and Western Saharan coasts since implementation of monitoring in 1999 (Taleb et al., 2006; Elgarch et al., 2008). In 2003 DSP toxins dominated by OA and DTX2 were detected in mussels, clams and oysters extending from El Jadida to Dakhla. In 2006 the presence of DSP toxins was investigated in Oualidia lagoon using liquid chromatography-mass spectrometry (LC-MS) and a quantitative enzyme-linked immunosorbent assay (ELISA) (Elgarch et al., 2008). Sampling from May to August showed OA concentrations to always exceed the regulatory level of $16 \mu\text{g } 100 \text{ g}^{-1}$ of edible tissue peaking in June at $135 \mu\text{g } 100 \text{ g}^{-1}$. DTX2 was also present particularly during July and August, contributing between 9 and 23% to the DSP toxins. Unfortunately the causative phytoplankton species were not identified.

In 2004 and 2005 mussels (*Mytilus galloprovincialis*) collected from Oulad Ghanem and Dar El Hamra on the Atlantic coast of Morocco and screened for lipophilic toxins by LC-MS were shown to contain DSP toxins and AZAs responsible for azaspiracid poisoning (AZP) (Taleb et al., 2006). This was the first report of AZAs in the CCLME and showed AZA2 to dominate with lesser quantities of AZA1 and traces of AZA3. In both 2004 and 2005 maximum concentrations of AZAs were recorded in summer and tended to follow peaks in DSP toxins dominated by OA and DTX2. The highest concentration of AZAs of $0.9 \mu\text{g g}^{-1}$ of digestive gland was recorded in mid-July 2005. AZAs were again detected in 2006, occurring on this occasion in Oualidia lagoon in July and August (Elgarch et al., 2008). On no occasion were the toxins of AZP found to exceed regulatory limits.

Water discolourations caused by blooms of *L. polyedrum* are well known on the Atlantic coast of Morocco (Tahri Joutei et al., 2000) and are consistent with reports of the dominance of *L. polyedrum* cysts in recent sediments (Targarona et al., 1999). Despite the prominence of *L. polyedrum*, YTXs have not been explicitly reported within the Northwest African Upwelling System (NW-AUS). However, following a monospecific bloom of *L. polyedrum* in July and August 1999, shellfish tested positive by means of the mouse bioassay for lipophilic toxins, indicating in all likelihood the presence of YTXs.

4.5.2.3. Domoic acid and amnesic shellfish poisoning

Amnesic shellfish poisoning (ASP) is caused by the neurotoxin domoic acid (DA) produced by some species of the globally distributed diatom genus *Pseudo-nitzschia* (Lelong et al., 2012). The impacts of *Pseudo-nitzschia* and its toxin have been considered by Trainer et al. (2010) to be especially problematic in upwelling systems, particularly in the California Current system where *P. multiseries* and *P. australis* have been identified as posing the greatest risk. Within the Iberian upwelling system, DA was first detected in Galician mussels in 1994, at which time *P. australis* was shown to dominate the plankton and significant amounts of DA were found in concentrated plankton samples (Míguez et al., 1996). This report provided the first account of DA in shellfish in Europe and the first record of *P. australis* in the North Atlantic Ocean.

The only study relating to ASP in the NW-AUS, was the identification by Akallal et al. (2002) of seven *Pseudo-nitzschia* species on the Atlantic coast of Morocco (*P. fraudulenta*, *P. multiseries*, *P. multistriata*, *P. pungens* var. *cingulata*, *P. subpacifica*, *P. delicatissima* and *P. pseudodelicatissima*), most of which have demonstrated the capability of toxin production. In addition to this study Rijal Leblad et al. (2013) have explored the diversity of *Pseudo-nitzschia* species and the potential risk of ASP in M'diq Bay on the western Mediterranean coast of Morocco in close proximity to the Gibraltar Strait and Atlantic Ocean. Here regular sampling during 2007 revealed the presence of 13 *Pseudo-nitzschia* species, eight of which are known DA producers. Elevated abundance of *Pseudo-nitzschia* spp. was recorded from March through to November and DA concentrations in shellfish were highest in spring, but did not exceed regulatory limits.

4.5.2.4. Ciguatera fish poisoning

Ciguatera fish poisoning (CFP) is a seafood-borne illness caused by consumption of fish that have accumulated lipid-soluble, polyether toxins known as ciguatoxins (CTXs) which have their origin in benthic dinoflagellates of the genus *Gambierdiscus* (Dickey and Plakas, 2010). The microalgae are ingested by herbivorous fish and assimilated and metabolized through multiple trophic levels of the food web. The illness is typically associated with the consumption of carnivorous fish, from tropical and subtropical areas of the Pacific and West Indian Oceans and the Caribbean Sea, producing a complex array of gastrointestinal, neurological and cardiological symptoms.

The CCLME was not considered a ciguatera-endemic region until an incident of poisoning in the Canary Islands in January 2004 provided the first indication of CFP in the northeast Atlantic. The incident followed consumption by five family members of an amberjack (*Seriola rivoliana*) caught by scuba divers off the coast of the Canary Islands resulting in illness with symptoms of CFP (Pérez-Arellano et al., 2005). A portion of the fish tested positive for CTXs using a commercially available immunoassay (Cigua-Check Test Kit). A further sample of the fish was sent to the Food and Drug Administration (FDA) Gulf Coast Seafood Laboratories (USA) and results were positive indicating a CTX content of 1.0 ng eq g⁻¹ as determined by a sodium channel-specific cytotoxicity assay. The toxin CCTX-1 was confirmed by LC-MS/MS and at least 2 other toxins with CTX-like structures were identified.

Further cases of CFP were investigated in the Canary Islands in 2008 and 2009 (Boada et al., 2010). In both cases symptoms followed consumption of amberjack, as in 2004. Toxin activity in both years was detected by cytotoxicity assay, at a level of 0.17 ng eq g⁻¹ in 2008 and 0.08 ng eq g⁻¹ in 2009. Again LC-MS/MS was used to confirm the presence of CCTX-1 in both years. Following these outbreaks a Surveillance System for Ciguatera poisoning in the Canary Islands (SVEICC) was established in 2009 requiring compulsory notification of all cases of suspected CFP in the healthcare system (Núñez et al., 2012). Between 2008 and 2012 a total of 9 CFP outbreaks have been reported in the Canary Islands affecting 68 people. All cases have been associated with the consumption of large amberjack, but in only three cases has the presence of CTXs been confirmed (Table 4.5.1).

Table 4.5.1. Ciguatera fish poisoning outbreaks off the Canary Islands from 2008-2012 (from Núñez et al., 2012)

Outbreak no.	Date	Island	No. human cases	Fish species	Weight	Origin
1	15 Nov 08	Tenerife	25	<i>Seriola fasciata</i>	37	Local market
2	29 Jan 09	Tenerife	4	<i>Seriola dumerilis</i>	67	Sport fishing
3	03Sep 09	Gran Canaria	3	<i>Seriola</i> spp.	Unknown	Unknown
4	19 Nov 09	Tenerife	2	<i>Seriola</i> spp.	Unknown	Sport fishing
5	25 Apr 10	Tenerife	6	<i>Seriola</i> spp.	80	Unknown
6	26 Jun 11	Gran Canaria	5	<i>Seriola</i> spp.	24	Sport fishing
7	28 Jan 12	Lanzarote	10	<i>Seriola</i> spp.	15	Sport fishing
8	04 Apr 12	Lanzarote	9	<i>Seriola</i> spp.	26	Sport fishing
9	May 12	Tenerife	4	<i>Seriola</i> spp.	Unknown	Local market

In 2007 and 2008 suspected cases of CFP were also recorded from the Madeira archipelago following consumption of fish caught off the Selvagens Islands (Otero et al., 2010). The 2008 incident affected 11 crew members of a fishing boat having caught and eaten amberjack (*Seriola* spp.). Their hospitalization and lengthy recovery period led to a clear diagnosis of CFP. Owing to the implication of *Seriola* spp. in these cases of CFP, two species, *S. dumerili* and *S. fasciata*, were caught off the Selvagens Islands in 2009 for analysis of CTXs. Assays using the Cigua-Check Test Kit provided positive results for both fish species. Ultraperformance liquid chromatography–mass spectrometry (UPLC-MS) revealed the presence of 4 CTX analogues, 2 of which were identified as CTX-1B and CTX-3C. The total toxin concentration as measured by UPLC-MS was 35 ng g⁻¹ in *S. fasciata*, and ranged from 33-54 ng g⁻¹ in *S. dumerili* depending on the tissue type.

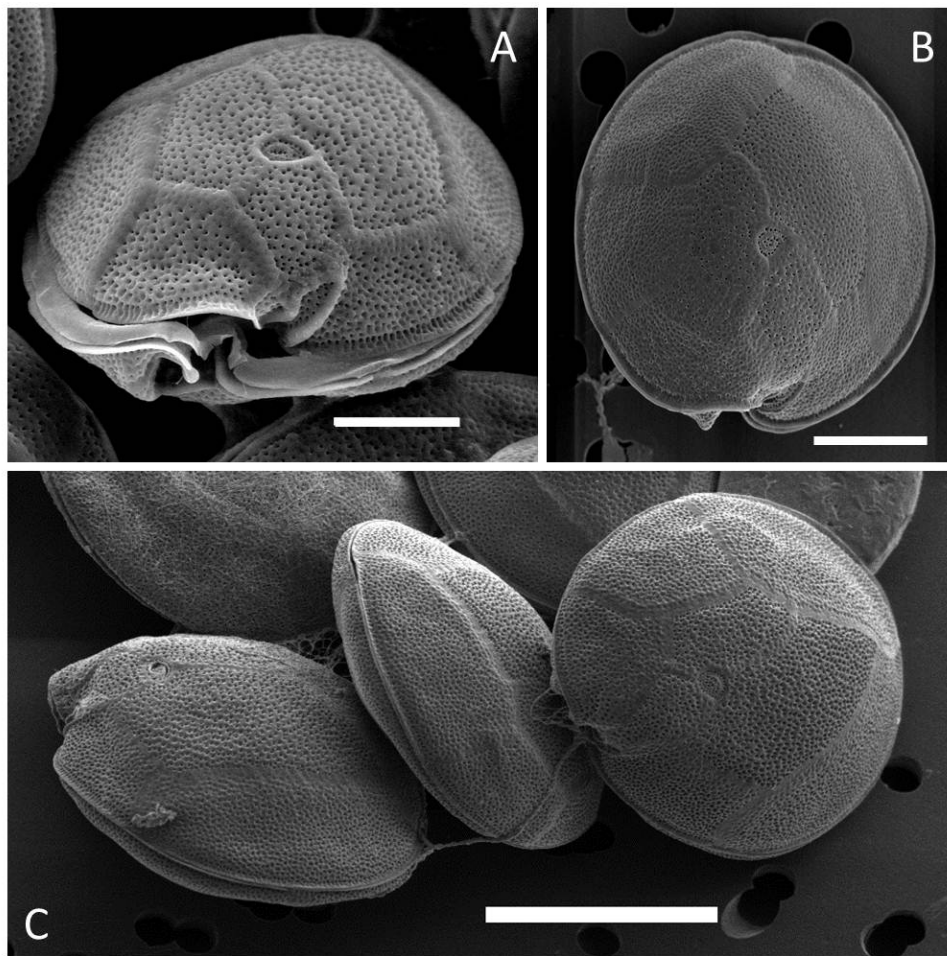


Figure 4.5.2. Three *Gambierdiscus* spp. found to co-occur in tidal pools of rocky shores of the Canary Islands: (A) *G. australes*, (B) *G. silvae* and (C) *G. excentricus* (scale bar = 20 µm; photographs by S. Fraga).

In attempting to establish the causative species of CFP in the Canary Islands, three *Gambierdiscus* spp. have been identified: *Gambierdiscus australes*, *Gambierdiscus excentricus* and *Gambierdiscus silvae* (Figure 4.5.2) (Fraga et al., 2011; Fraga and Rodríguez, 2014). Although the northeast Atlantic was not considered a ciguatera-endemic zone prior to 2014, it has been suggested by Fraga et al. (2011) that the oldest record of CFP may date back to 1521 in the Gulf of Guinea, and that the first recorded observation of *Gambierdiscus* was in all likelihood made from the Cape Verde archipelago in 1948. Reported at the time as *Goniodoma* sp. by Silva (1956) this species is most likely the same as that recently described as *G. silvae* by Fraga and

Rodríguez (2014). Found as an epiphyte on small seaweeds in tidal pools on the northeast coasts of the islands of Gran Canaria and Tenerife, phylogenetic analyses show strains of *G. silvae* to cluster in a sister clade to *G. polynesiensis*. Previously Fraga et al. (2011) had also described the species *G. excentricus* from the Canary Islands. Shown by phylogenetic analysis to cluster into a well-supported group, with *G. australes* its closest relative, all strains of *G. excentricus* were shown to be toxic indicating the production of both CTX and maitotoxin (MTX)-like compounds. Cell content of CTX was found to range from 0.37-1.1 pg CTX-1B eq cell⁻¹, similar to that reported previously for other species of *Gambierdiscus*. However, MTX content ranged from 0.48-1.38 ng cell⁻¹, classifying *G. excentricus* as a potent MTX producer. The work of Fraga and Rodríguez (2014) further provided the first identification of *G. australes* in the Atlantic, which was found to co-occur with *G. excentricus* and *G. silvae*.

These reports of *Gambierdiscus* spp. and ciguatoxic fish in the Canary Island and Madeira archipelagos are of further significance in providing evidence of biogeographical expansion, with a shift towards higher latitudes, which has been considered indicative of climate change and increasing sea temperatures in the region (Dickey and Plakas et al., 2010, Otero et al., 2010).

4.5.2.5. Toxin producing cyanobacteria blooms

Several cyanobacteria occurring in fresh, brackish and marine waters produce toxins which are hazardous to the health of livestock and humans. The toxins are classified, according to the target of their toxic action, and include primarily hepatotoxins, neurotoxins and dermatotoxins. Cyanobacteria blooms in marine waters are limited to only a few taxa with *Trichodesmium*, *Richelia*, *Nodularia* and *Aphanizomenon* being most commonly observed (Sellner, 1997).

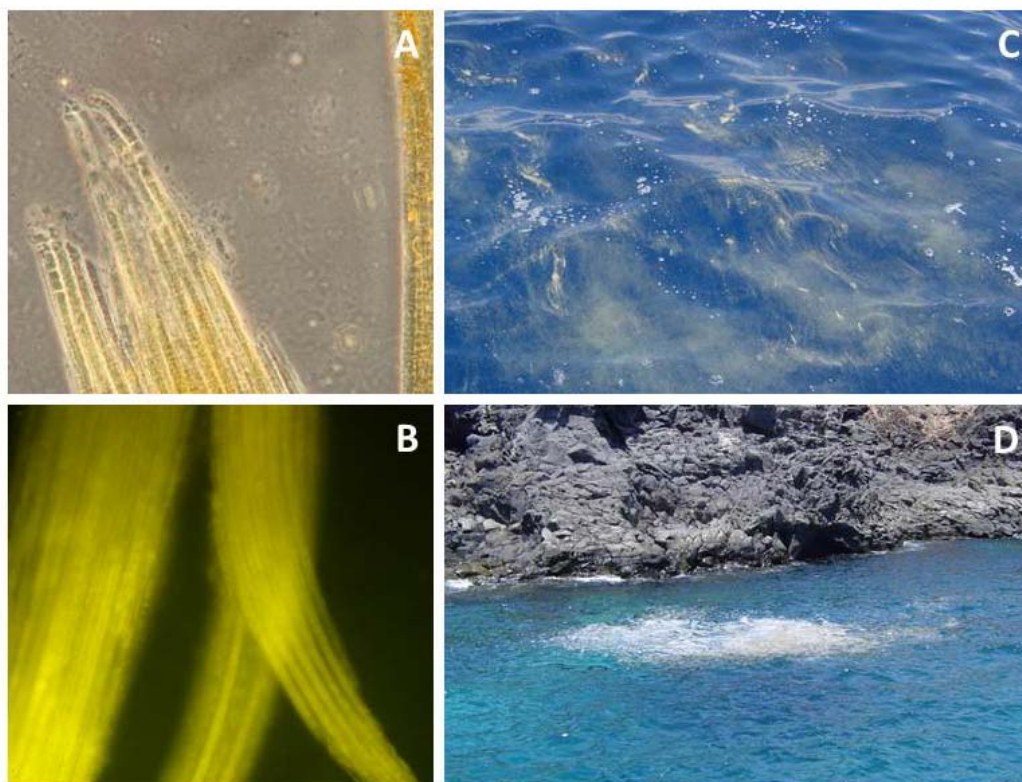


Figure 4.5.3. *Trichodesmium erythraeum* bloom in the Canary Islands in August 2004. Light micrographs of filaments using (A) phase and (B) fluorescence microscopy. Mats of surface filaments (C) and surface foam created by these mats (D) adjacent to the rocky shores of Tenerife (photographs A, B by S. Fraga and C, D by M. Norte).

In August 2004 an extensive bloom of the cyanobacterium *Trichodesmium erythraeum* was observed in the NW-AUS (Ramos et al., 2005). Water samples collected off the islands of Gran Canaria and Tenerife were found to contain as many as 1240 filaments ml⁻¹ of this non-heterocystous diazotrophic cyanobacterium, and in some cases were observed to form filamentous mats in the waters surrounding the islands (Figure 4.5.3). Microcystins were detected by HPLC and confirmed by immunoassay at concentrations from 0.1–1.0 µg microcystin-LR eq g⁻¹ dry weight of bloom material. The bloom corresponded to the warmest summer in the Canary Islands since 1912 and followed severe dust storms in the Sahara Desert. Dust deposition is considered to extend over considerable areas of the northeast Atlantic with a significant impact on the biogeochemical cycle of trace elements, specifically providing a source of Fe required for the growth of diazotrophic cyanobacteria. These are considered the first records of extensive *T. erythraeum* blooms off the NW-AUS and may be a function of a general warming of the area (Ramos et al., 2005).

4.5.3. CONCLUSION

Knowledge of HAB species and their impacts in the major EBUS is disparate between systems and is least studied off NWA. Nevertheless the few documented studies of HABs within this region indicate a similar diversity to that recorded in other upwelling systems, and include those species responsible for PSP, DSP, ASP and AZP. Also present off NWA, but generally absent from the other major upwelling systems, are those species responsible for CFP and microcystin-producing cyanobacterial blooms. Their presence is afforded by the subtropical habitat provided by the island archipelagos found within the CCLME. It is intended that this brief review will provide the foundation and stimulus for further studies of the ecology and dynamics of HABs, of their toxins, and of the public health and socioeconomic impacts of HABs within this region.

Acknowledgements

We thank the Intergovernmental Oceanographic Commission of UNESCO for the invitation to contribute this article through participation in the workshop on *Oceanographic and Biological Features and Trends in the CCLME* in Las Palmas de Gran Canaria from 27-29 January 2015.

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